In vitro propagation of *Galtonia* species

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Leaf, gynoecium, stigma + style, ovary, pedicel and peduncle explants were excised from *Galtonia candicans* (Bak.) Decne and *G. viridiflora* Verdoorn plants and placed on Murashige and Skoog medium supplemented with 1 mg l^{-1} NAA and 0.3 mg l^{-1} BA. Shoots were initiated on the pedicels of *G. candicans* and at the base of the ovary of *G. viridiflora* explants. Prolific shoot production resulted when these shoots were cut into quarters and placed on fresh medium. In the absence of BA, addition of only 0.5 mg l^{-1} NAA to the basic medium resulted in rooting of the shoots. The plantlets were then successfully hardened off in a peat:sand (1:1) (v/v) mix.

Blaar-, stamper-, stempel + styl-, vrugbeginsel-, blomsteel- en bloei-as- eksplante is verkry van *Galtonia candicans* (Bak.) Decne en *G. viridiflora* Verdoorn plante en geplaas op Murashige-en-Skoog basiese medium wat 1 mg l^{-1} NAA en 0.3 mg l^{-1} BA bevat het. Lootjies het op die blomsteel-eksplante van *G. candicans* en aan die basis van die vrugbeginsel-eksplante van *G. viridiflora* gevorm. Oorvloedige lootvorming het plaasgevind toe die gevormde lootjies in kwarte verdeel en op vars medium oorgeplaas is. In die afwesigheid van BA het die byvoeging van 0.5 mg l^{-1} NAA by die basiese medium wortelvorming by die lootjies tot gevolg gehad. Die gevormde plantjies is suksesvol in 'n veen:sand-mengsel (1:1) (v/v) afgehard.

Keywords: *Galtonia candicans, Galtonia viridiflora,* propagation, tissue culture.

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Galtonia candicans (Bak.) Decne and *G. viridiflora* Verdoorn are South African members of the Hyacinthaceae/Liliaceae. Both are found in mountainous country, at altitudes of between 1500 and 2200 m (Batten 1986). *G. candicans* has beautiful large, pendulous, creamy-white flowers (Gibson 1978), whereas those of *G. viridiflora* are decidedly green (Hilliard & Burtt 1988). The inflorescence of *G. candicans* extends from the leaves and can reach a height of 1.5 m (Gibson 1978). Each bulb produces several inflorescences in succession, thus prolonging the flowering season (Batten 1986). *G. candicans* is apparently easily grown from seed (Batten 1986; Hilliard & Burtt 1988), but local nurserymen report difficulty in propagating these two *Galtonia* species vegetatively.

The objective of this study was therefore to propagate *Galtonia in vitro*. *G. candicans* has been propagated by twin scaling (Sakanishi & Yanagawa 1979). We chose, however, to use explants other than bulb material in order not to destroy the mother plant. The explants used included the gynoecium, stigma + style, ovary, pedicel, peduncle and leaf. The floral parts were detached from the parent plant five days after anthesis. The limited availability of these explants meant that replicates of different treatments or different explant types were restricted and variable.

All explants were sterilized by dipping them in ethanol for five seconds, followed by a 15-min soak in 0.5% sodium hypochlorite. Thereafter, they were rinsed four times in sterile distilled water, for 10 min at a time. The flowers were dissected in various ways to produce a variety of explants. These included: the flower with the petals removed and the pedicel inserted into the agar medium; segments of the pedicel inserted into the medium; pedicel segments split in half and placed onto the surface of the medium; ovaries cut in half vertically and placed onto the medium; stigmas with styles inserted into the medium; and peduncle segments placed onto the medium. In addition, 1-cm^2 leaf explants were placed onto the medium.

All explants were placed onto media in 25 × 100-mm test tubes closed with Cap-O-Test lids and sealed with parafilm, or in glass screw-top jars, in the case of large explants. The cultures were maintained in a culture room at 25 ± 2°C under a low light intensity of 0.5 μmol m^{-2} s^{-1}. Media formulations listed by George *et al.* (1987) for the culture of *Ornithogalum*, a related species, were used as a guideline for the culture of *Galtonia*. The media tested were full- and half-strength Murashige and Skoog (1962) salts, with 0.5 mg l^{-1} thiamine hydrochloride, 0.1 g l^{-1} myo-inositol, 30 g l^{-1} sucrose, 1 mg l^{-1} 1-naphthaleneacetic acid (NAA) and 0.3 mg l^{-1} 6-benzylaminopurine (BA). The pH was adjusted to 5.8 and 8 g l^{-1} Unilab agar was added prior to autoclaving.

Within a few days, the ovaries of the intact flowers had abscised, leaving just the pedicel inserted into the agar. Contamination was not observed in any tube or bottle. After five weeks, little or no change was observed in most explants. However, shoots were visible on the pedicels of *G. candicans* from which the ovaries had abscised, both when half- and full-strength Murashige and Skoog (1962) salts (Figure 1) had been used. In addition, some callus and a shoot had formed at the base of one *G. viridiflora* ovary placed on half-strength Murashige and Skoog (1962) medium (Figure 2). No such response was evident in the other five explants. After ten weeks, no further organogenesis was observed in any *G. viridiflora* explants, but many of the pedicel segments of *G. candicans* inserted into the medium and those cut in half and placed onto the medium, had initiated shoots. This was most prevalent when half-strength Murashige and Skoog (1962) salts had been used. With time, the number of shoots gradually increased in both species, but in neither did any additional explants commence organogenesis.

Once the shoots were sufficiently large to be readily separated, some were cut into quarters and placed on fresh medium. Within one week, some shooting was observed, and within six weeks, shoots had been initiated on all explants. This was especially successful with *G. viridiflora*. The shoots that were not used to initiate new shoots, were placed on a rooting medium containing half-strength Murashige and Skoog (1962) salts, 0.5 mg l^{-1} NAA and no cyto-

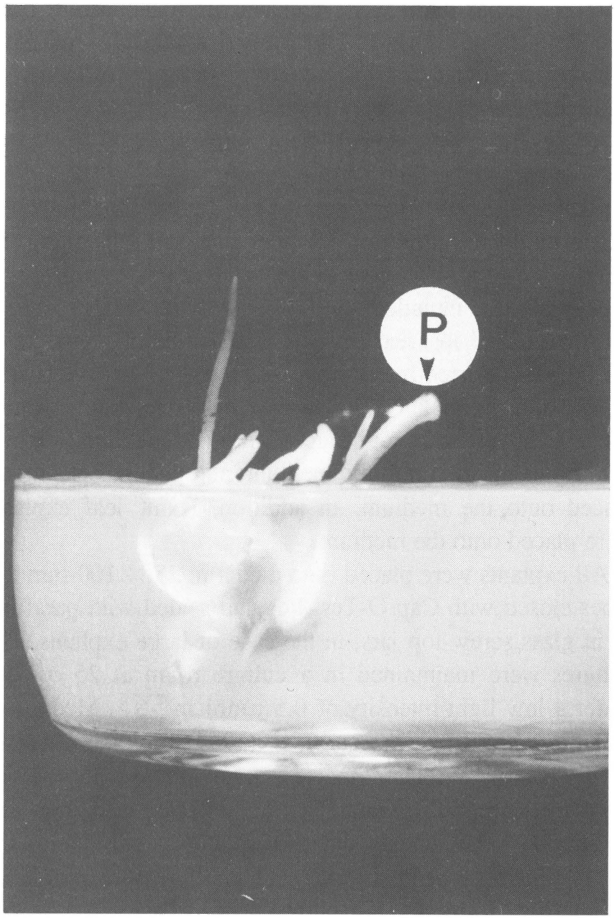

Figure 1 Shoot initiation from the pedicel (P) of a *G. candicans* flower, from which the ovary had abscised after three days. The medium contained 1 mg l^{-1} NAA and 0.3 mg l^{-1} BA.

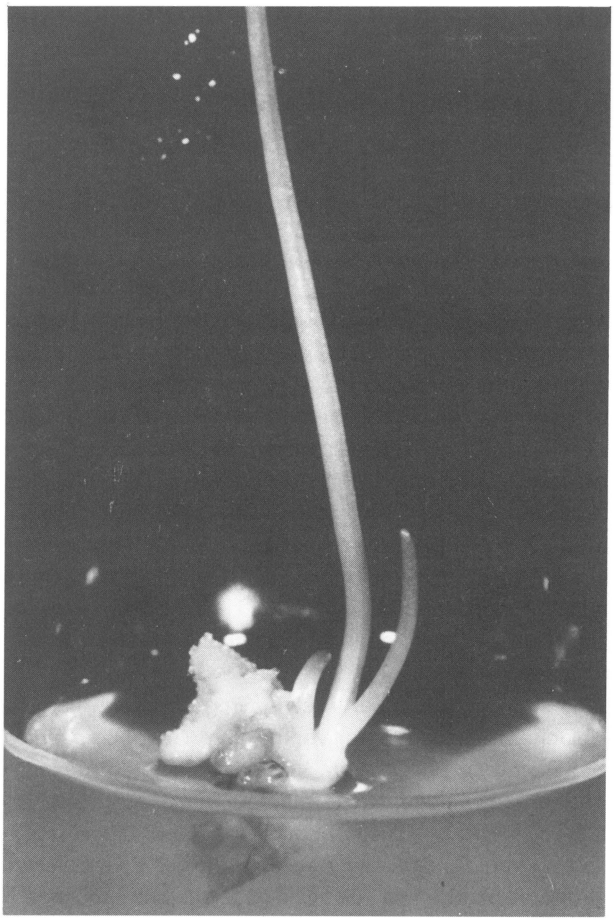

Figure 2 Shoot and callus initiation from a sectioned ovary of *G. viridiflora* placed on a medium containing half-strength Murashige and Skoog (1962) salts, 1 mg l^{-1} NAA and 0.3 mg l^{-1} BA.

kinin. Within three weeks, the shoots had rooted, and four weeks later, they were hardened off. This was achieved by planting the plantlets in an autoclaved peat:sand (1:1) (v/v) mix in a misthouse for two weeks. Thereafter, they were transferred outside under shadecloth. The survival rate was 95% for *G. viridiflora* (n = 17) and 82% for *G. candicans* (n = 106).

Overall, the most successful means of *in vitro* propagation was found to be the use of pedicel material for *G. candicans* and sectioned ovaries for *G. viridiflora*, followed by the sectioning and culture of shoots initiated in this way. This non-destructive technique has the additional advantage of involving explant material which is more readily sterilized than bulb material. Disadvantages include the fact that flowers are produced seasonally, but this is balanced by the possibility of continuous production of new shoots from the shoots initially produced in culture. By these means, approximately 1000 *G. viridiflora* and 600 *G. candicans* plantlets could be produced in nine months from a single initially responsive explant.

Acknowledgements

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